How to recognize and treat an infection with *Bsal*

The emerging infectious chytrid fungus *Batrachochytrium salamandrivorans* (*Bsal*) causes mass mortality events in both captive collections of salamanders and newts as well as in wild living populations of at least fire salamanders (*Salamandra salamandra*). Swift and accurate detection of the pathogen is of utmost importance to prevent further expansion of this pathogen. This leaflet provides veterinarians an overview of macroscopic and microscopic lesions, the required diagnostic tests to confirm diagnosis, and the proper treatment.

Typical lesions, although not pathognomonic, consist of multifocal epidermal erosions and ulcera, often characterized by a black margin. The extent and size of the lesions range from asymptomatic (at the onset of the infection) 1-2 mm circular and localized lesions to large skin ulcera affecting the whole body. Dysecdysis, anorexia and ataxia may be present. Ultimately the animal dies

Microscopy

Microscopy includes wet mount preparations, histology, and immunohistochemistry, and requires pieces of whole or shed skin.

Histology/histopathology reveals keratinocytes with eosinophilic necrosis and marginated nuclei at the periphery of the erosions/ulcerations. Within these keratinocytes (mostly colonial) thalli can be present.

Immunohistochemistry is used to stain the chytrid fungus (no distinction between *Bd* and *Bsal*).

Wet mounts may reveal the presence of motile zoospores.

PCR/qPCR

Real-time PCR is a sensitive method to show the presence of Bsal ante- and post-mortem and can be applied to skin swabs or skin samples.

The Bsal- and Bd species-specific duplex real-time PCR allows simultaneous quantification of both chytrid fungi in amphibian samples. When used as a post-mortem diagnostic tool, the detection limit should be 1.0 GE of Bsal to prevent false positives.

Molecular diagnostic tools should be used in conjunction with histology or histopathology and clinical signs, where applicable.

Treatment

Exposing infected amphibians to temperatures of 25°C for a 10-day period will result in clearance of infection and the healing of associated lesions. This is of course taking into consideration the clinical stage of the disease and the amphibians' thermal tolerance (many urodeles tolerate these relatively high temperatures poorly).

Alternatively: a treatment protocol of a combination of Voriconazole 12.5 µg/ml, Polymyxin E 2000 IU/ml at a temperature of 20°C clears the infection in infected salamanders in 10 days.

More information, literature, diagnostic and reference labs are available via: www.BsalEurope.com and Ghent University, Wildlife Health Ghent, Merelbeke (Belgium).

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